

Irradiation dependent inflammatory response may enhance satellite cell engraftment

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Supplementary data

Macro for quantifying TUNEL+ cells

Macros ran on FIJI. The DAPI count was written by Dale Moulding. The TUNEL count was adapted by B.D. for counting TUNEL positive cells.

DAPI count

```
dir1 = getDirectory("Input folder"); //select an input folder
dir2 = getDirectory("Choose a folder to save to"); //select an output folder.
list = getFileList(dir1); //make a list of the filenames
setBatchMode(true); //turn on batch mode so it runs in the background

    if (isOpen("Results")) {
        selectWindow("Results");
        run("Close");
    }

// repeats the macro for every file in the folder using a for loop.

    for (i=0; i<list.length; i++) {
        showProgress(i+1, list.length);
        filename = dir1 + list[i];

        open(filename);
        Imagename = File.nameWithoutExtension;

        // set the value for the Gaussians
        GausLow = 7;
        GausHigh = 10;
        run("Brightness/Contrast...");
        run("Enhance Contrast", "saturated=0.35");
        run("Duplicate...", "title=GausLow duplicate channels=3");
```

```

run("Duplicate...", "title=GausHigh");
run("Gaussian Blur...", "sigma="+GausHigh);
selectWindow("GausLow");
run("Gaussian Blur...", "sigma="+GausLow); // set the value for the low gaussian
imageCalculator("Subtract create", "GausLow", "GausHigh");
selectWindow("Result of GausLow");
run("Find Maxima...", "prominence=15 output=Count");
setResult("Image", i, Imagename);
updateResults();
selectWindow("Result of GausLow");
run("Find Maxima...", "prominence=15 output=[Point Selection]");
roiManager("Add");
selectWindow("GausLow");
close();
selectWindow("GausHigh");
close();
number=roiManager("count");
roiManager("select", number-1);
roiManager("Rename", "Dapi_"+Imagename);
run("Close All");
}

```

```

roiManager("Save", dir2+"Nuclei-RoiSet.zip"); // saves the ROIs .

```

```

selectWindow("Results");

```

```

saveAs("Results", dir2+"Nuclei-Counts.csv"); // save the summary as a .csv.

```

```

exit("Nuclei measured in "+i+" images"); // close the macro and display a window with
number of images processed.

```

TUNEL Count

```

dir1 = getDirectory("Input folder"); //select an input folder

```

```
dir2 = getDirectory("Choose a folder to save to"); //select an output folder.
```

```
list = getFileList(dir1); //make a list of the filenames
```

```
setBatchMode(true); //turn on batch mode so it runs in the background
```

```
    if (isOpen("Results")) {  
        selectWindow("Results");  
        run("Close");  
    }
```

```
// repeats the macro for every file in the folder using a for loop.
```

```
    for (i=0; i<list.length; i++) {  
        showProgress(i+1, list.length);  
        filename = dir1 + list[i];
```

```
        open(filename);  
        Imagename = File.nameWithoutExtension;  
        imagename= File.name();
```

```
        // set the value for the Gaussians
```

```
        GausLow = 2.75;
```

```
        GausHigh = 5;
```

```
        run("Split Channels");
```

```
        selectWindow(imagename+" (green)");
```

```
        run("Duplicate...", "title=GausLow duplicate channels=3");
```

```
        run("Duplicate...", "title=GausHigh");
```

```
        run("Gaussian Blur...", "sigma="+GausHigh);
```

```
        selectWindow("GausLow");
```

```
        run("Gaussian Blur...", "sigma="+GausLow); // set the value for the low gaussian
```

```
        imageCalculator("Subtract create", "GausLow", "GausHigh");
```

```

selectWindow("Result of GausLow");

run("Find Maxima...", "prominence=30 output=Count");

setResult("Image", i, Imagename);

updateResults();

selectWindow("Result of GausLow");

run("Find Maxima...", "prominence=30 output=[Point Selection]");

roiManager("Add");

selectWindow("GausLow");

close();

selectWindow("GausHigh");

close();

number=roiManager("count");

roiManager("select", number-1);

roiManager("Rename", "TUNEL_"+Imagename);

run("Close All");

}

```

```

roiManager("Save", dir2+"TUNEL-RoiSet.zip"); // saves the ROIs .

```

```

selectWindow("Results");

```

```

saveAs("Results", dir2+"TUNEL-Counts.csv");// save the summary as a .csv.

```

```

exit("TUNEL measured in "+i+" images"); // close the macro and display a window with
number of images processed.

```

Supplementary Table 1. GSEA results for enriched genesets with an FDR value below 0.05, with a large amount of gene ontology enrichments related to the activation of an innate immune and inflammatory responses.